بسم الله الرحمن الرحيم

INVESTIGATION OF PARASITES OF WORKING DONKEYS IN KHARTOUM STATE, SUDAN

By

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DEDICATION

To the soul of my lovely sisterAsma
To my lovely father
To my mother
To my husband
To my daughter Asma
To my son Ahmed
To my relatives
For those who were in my heart and are still there,
thank you

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ABSTRACT

This study was conducted in Khartoum State on apparently healthy donkeys used as draft animals. It involved one hundred donkeys. Thirty five blood and feacal samples were taken from Omderman,35 from Khartoum North and 30 from Khartoum , the samples of faecal material were examined for worms load and faecal cultures were studied. Blood samples were taken for parasitic examination and were injected into mice as laboratory animals to find out the possibility of the presence of blood parasites. The results of fecal culture showed the existence of different kinds and degrees of infection with nematodes in Omdurman, Khartoum North and Khartoum. The Nematodes rates were found to be 65.5%, 60.8% and 56.6% in each locality respectively. Nematodes involved were *Srongylus spp* and *Trichonema spp*, worm eggs were found in great numbers. The blood samples did not disclose any blood parasites. When the samples were Injected into the mice and examined for a month no blood parasites were detected. Donkeys examined for parasite worms showed heavy parasite burden but with no blood parasites. When examined for ticks the donkeys were found clean of ticks. The reason could be that the donkeys owners injected their animals with Ivermectin regularly.

المستخلص

أجريت هذه الدراسة في ولاية الخرطوم على مائة حمار تـستخدم لنقل الاحمال كانت سليمه ظاهرياً اخذت 35 عينة دم وبراز من عـدد 35 حمار من ام درمان و35 من الخرطوم بحري و30 من الخرطوم.

فحصت عينات البراز لمعرفة الديدان الموجـوده فيهـا وأخـذت عينات من دمها ومن ثم حقنت في فئران تجارب لمعرفة أمكانية وجـود طفيليات دم فى دم الحمير.

أشارت نتائج زراعة البراز إلي وجود ديدان النيمـاتودا بـانواع مختلفة ومعدلات مختلفة في كل مـن ام درمـان والخرطـوم بحـري والخرطوم بنسب 65.5% ، 60.8% ، 56.6% في كل منطقـة علـى التوالى وشملت النيماتودات أنواع الاسترونقاياس والترايكونيما، أما بيض الديدان فقد وجد بإعداد كبيرة. لم تظهر عينات دم الحمير وجود لطفيليات الدم. وعندما حقنت فئران التجارب بعينات الدم هذه وأجريت فحوصات لدم الفئران لمدة شهر لم تظهر أي طفيليات دم. وعندما فحصت الحمير للقراد وجدت انها نظيفه من القراد ربما يعزي السبب إلي أن اصـحاب الحمير يداومون على إعطائها ايفرمكتين ضد الطفيليات بـصورة شـبه منتظمة.

CHAPTER ONE

INTRODUCTION

Despite the increase in mechanization throughout the world donkeys still remain well deserving the name ‘beasts of burden'. They have prominent position in the agricultural activities of many developing countries. This is shown by the widespread use of donkeys in rural and urban areas in Africa (Peason Missed*l*., 1999). It is suggested that donkeys continue to play a great role in farm work and contribute to food security and social welfare of many impoverished communities.

Even though donkeys have often been described as sturdy animals, they succumb to a variety of diseases (Svendsen, 1997). The attention given the donkey has always been below to what it deserves. This might be partly due to the wrong perception that the donkey does not require a lot of care when they get sick and the donkeys are of low social status.

Despite the huge contribution of donkeys to improvement of human life their health problems affecting their welfare are neglected in most parts of the country.

In the Sudan the donkey belongs to the African wild ass *Asinus afreicanus* (Hatenorth and Diller, 1986).

In the semi-desert regions of Darfur and Eastern Chad donkey is not just abeast of burden. It is a living pickup truck carrying water of firewood. Refugee family loads their days supply of water onto donkey are also needed to carry precious firewood into the camps (Scott, 2007).

There is very little information available about donkey types and their distribution in the Sudan. A report from the Red Sea Province showed the principal types of the donkey there covering the indigenous varieties in the Province. The report described them according to their phenotypic characters and referred them to the African wild ass *Assinus africanus sub sp africanus* and *faenopus* (Hatenorth and Diller, 1986). The breed and types encountered in this province, according to the report were Makadi and Makadawi and their dominant varieties are Hamshawi and Refawi.

Their origins are Eldamer and Atbara in the north and Galabi from Gezira in the central Sudan. The number and distribution of donkeys and horses in Sudan is given by the Statistical Bulletin for Animal Resources and is shown in table (1).

Table (1): Number and distribution of donkey in the Sudan

State	Horses	(%)	donkeys	(%)
North Kordofan	69000	10.6	635000	10.0
South Kordofan	13000	2.0	273050	4.3
West Kordofan	43000	6.6	571500	9.0
North Darfur	16000	2.5	673100	10.6
South Darfur	225000	34.6	514350	8.1
West Darfur	69000	26.0	774700	12.2
Elgedarief	13000	1.5	746250	4.3
Kassala	8000	1.2	273050	4.3
Red Sea	10000	1.5	196850	3.1
Blue Nile	13000	2.0	228600	3.6
Sennar	10000	1.5	247650	3.9
Elgezira	22000	3.4	768350	12.1
White Nile	28000	3.4	393700	6.1
Northern	1000	0.2	146050	2.3
River Nile	2000	0.3	158750	2.5
Khartoum	8000	1.2	6350	0.1
North Upper Nile	500	0.1	6350	0.1
Unity	2000	0.3	0	0
Gongoli	0	0	0	0
N.Bahr elGhazal	500	0.1	6350	0.1
Bahrel Ghazal	0	0	0	0
Albohairat	0	0	0	0
Warab	0	0	0	0
Bahar Elgabal	0	0	0	0
E. Equatoria	0	0	0	0
W. Equatoria	0	0	0	0
Total	650000	100.0	6350000	100%

*From Statistical Bulletin for Animal Resources 2000, Animal Resources. Issue No. (10) p. 75.

A single donkey weighing 120-180 kg is capable of pulling draft load of about 20-30 kg for several hours. Greater force in excess of 40% of body weight can be exerted for short periods (Betker and Kutzbach,H.D, 1991).

Parasites are some of the important causes of reducing the donkey ability to work and some times lead to animal loss. Donkey parasites are of two types: internal parasites and external parasites. The donkeys are animals which suffer from parasites for a long time without getting adversely affected except for their getting weak.

Objectives:

To determine the prevalence of parasites in donkeys apparently healthy male draft in Khartoum State during March, 2008

CHAPTER TWO

LITERATURE REVIEW

2.1 Classification of donkeys(Carl Van Linne 1775)

Kingdom	:	Animalia
Phylum	:	Chordata
Class	:	Mammalia
Order	:	Perissodactyla
Family	:	Equidae
Genus	:	Equus
Subgenus	:	Asinus
Species	:	Equus asinus
Sub-species	:	Equuasinus africanus Equus asinus somalicus

2.2 Donkeys population and history

The donkey or ass (*Equus asinus*) is originated from wild asses in Africa and Asia. In Africa there are two different species the Nubian from the north and the Somali from Far East to south of Red Sea. The ancestors of the modern donkeys are the Nubian and Somalian sub-species of African wild ass which was domesticated around 4000 B.C. The donkey became an important pack animal for people living in the Egyptian and Nubian regions as they can easily carry 20-30% of their

own body weight and can also be used as a farming and dairy animal. By 1800 BC the ass had reached the Middle East where the trading city of Damascus was referred to as the (city of asses) (Clutton, 1999).

For the Greeks the donkey was associated with the Syrian God of wine Dionysus. The Romans also valued the ass and used it as a sacrificial animal.

Equines had become extinct in the Western Hemisphere at the end of the last Ice Age. However, horses and donkeys were brought back to the Americas by the conquistadors in 1495. The ass first appeared in the New World when Christopher Columbus brought four jacks and two jennys there (Malcolm Starkey, 2003).

Shortly after the Americans became independent President George Washington imported the first mammoth jack stock into the country from Spain and France (Tylor *et al*., 2005).

Donkeys were found useful as pack animals by miners particularly the gold prospectors of the mid 1800s. Miners preferred this animal due to its ability to carry tools, supplies and ore, and their sociable disposition and adaptation to human companionship allowed many Miners to lead

their donkeys without ropes; they simply followed behind their owner (Clutton, 1999).

By the early 20th century donkeys began to be used less as working animals and instead were kept as pets in the United State and other wealthier nation, while remaining an important work animal in many poorer regions (Blench, 2000).

The domestication and historical development of the donkey are ancient knowledge obtained through archaeological and linguistic association. The donkey is indigenous to the Africa continent and its wild progenitor is usually considered to be Nubian wild ass, historically a chain of races of wild asses spread from the Atlas Maintains to the Red Sea and probably as far south as the border of present day Northern Kenya.

Wild asses have been domesticated as seen in Egypt in the fourth millennium BC.

The estimated donkey population of the world ranged between (30-45 million) and tended to increase (FAO, 1995).

At the year 2000 the number of donkeys in the Sudan was estimated to be more than (6 million). In Khartoum State in 1992 there

were more than 26000 and increased to more than 27000 in 1997; their number was to be more than that of horses in Khartoum State (Statistical Bulletin for Animal Resources, 2000 SBAR)

2.3 Description of the donkey

Colour in the donkey ranges from gray to brown. Donkeys come in a variety of sizes from the miniature Mediterranean (under 36 inches) to the elegant Mammoth Jackstock (14 inches and up), to the rare French piteous donkey characterized by its huge head and ears and very thick, shaggy, curled and black coat which can stand 14-15 hands high.

There are fewer than 200 purebred piteous left in the world. To day the types of the donkey are labeled by their size (Mason, 1988).

Wild donkeys live spreated from each other, unlike tight wild horses and feral horses herds, donkeys have developed very loud vocalizations which help keep in contact with other donkeys over the wide spaces of the desert. The best known call is referred to “bray” which can be heard for over three kilometers.

Donkeys have larger ears than horses, their longer ears may pick up more distant sound and may help cool the donkeys blood. Donkeys in

the wild can defend themselves with a powerful kick of their hind legs as well as by biting and striking with their front feet.

Donkeys tough digestive system is some what less prone to colic than that of horses. It can breakdown near-inedible vegetation and extract moisture from food very efficiently.

As a rule donkeys need smaller amount of feed than horses with comparable height and weight. If overfed, donkeys are also quite susceptible to developing a condition called laminitis.(Coma,2000)

2.4 Uses and importance of the donkey

Donkeys have been used throughout history for transportation of supplies pulling wagon and in a few cases as riding animals. During World War 1 a British stretcher bearer John Simpson serving with Australian and New Zealand army corps used a donkey to rescue wounded soldiers, carrying them to the safety places. Donkeys were until recently used in the Italian Army. The Mountain fusiliers each had a donkey to carry their gear and in extreme circumstances the animal could be eaten. In 2006, security forces in Afghanistan prevented a man taking a donkey which he had laden with 30 kg of explosives and a number of

landmines which would had been set off by a remote controlled detonator from entering the town in Zabul Province.

The donkey for his size is an excellent pack animal and it is so employed in many countries including Sudan, Egypt, Somalia and China. Its pace is low compared with the mule and may take a load of more than 30 kg. It will do well on poor grass or forage and is available for transport on a line of communication. Donkeys are useful pack animals if they are not over loaded or over driven, their pace is two miles and half an hour and they cover fifteen miles a day (Catley ,1995).

Donkeys are kept in Africa for four reasons: work, breeding, milking and eating. Of these work is by far the most important, donkeys are used mainly as pack animals either for carrying load or for riding, in arid regions. They are used together with camels to pull water from deep wells. Less commonly they are used to pull carts or plows. More than 90% of the estimated 44 million donkeys are present in developing countries doing work. Two million people depend on and use donkeys daily for transportation and for agricultural purposes (Starkey,et al 1997; FAO, 1995).

Breeding donkeys can be profitable business in certain regions of the Sahel. It is often more convenient for donkey users in the south to buy such animals from further north and replace them at the end of their working lives. Countries such as Nigeria and Mali have considerable trade selling donkeys usually males, to communities further south. The milking of donkeys in Africa is rare and of little economic importance however the western Massai are reported to milk donkeys (Starkey,et al 1997).

The world donkeys have been hunted to near extinction for meat and eating donkeys is common in many Eurasian pastoral systems.

The importance of donkey is steadily increasing in Sudan as well as many other African countries due to poverty. It plays an important role in the provision of energy for agricultural production by way of traction of cultivation and transport of products (Starkey,et al 1997).

Outside the major agricultural area it is used for carrying water. In rural area male donkey is preferred for both riding and transport. In town many people depend on it for their livelihood and is often seen harnessed to two wheel carts especially in market centers (Hatenorth and Diller, 1980).

2.5 Donkeys hybrids

A male donkey jack can be crossed with female horses to produce a mule; a male horse can be crossed with a female donkey to produce a hinny (American Donkey and Mule Society,).

Horse-donkey hybrids are almost always sterile because horses have 64 chromosomes whereas donkeys have 62 chromosomes, producing offspring with 63 chromosomes. Mules are much more common than hinnies, this is believed to be caused by two factors the first when the chromosome count of the male is higher fertility rates drop. The lower progesterone production of the hinny may also lead to early embryonic loss, in addition there are less scientific reasons due to different mating behavior. Zebras in captivity are most valuable when used to produce full-blooded zebras. There are not enough female zebras breeding in captivity to spare them for hybridizing and there is no such limitation on the number of female donkeys breeding (Burnham, 2002).

An animal which may look like a zebra-donkey hybrid because of its distinctly striped hindquarters and hind leg is the okapi which has no relationship to either of those species. Okapi are most closely related to

the giraffe in addition to rear stripes; okapi have some striping near the top of their forelegs.

2.6 Diseases which affect donkeys

Many of the diseases of the donkeys (and horses for that matter) have not been well characterized and so they are given the names of similar disorders in human and other animals (Fescha, 1997).

2.6.1 Colic

The term is applied to symptoms of abdominal pain. Donkeys are more stoical than the horse and symptoms are less violent.

There are different types of colic. The main types are spasomodic – thromboembolic, flatulaerd, impaction, obstruction and grass sickness (In Sudan the syndrome is frequently encountered veterinary clinic (Hamid *et al*., 1998; Aradaib and Abbas, 1985).

2.6.2 Leshmaniasis

The majority of leshmania infections are zoonatic in origin. The disease is transmitted by the sand fly. It occurs in 3 clinical forms, cutanous muco-cutaneous and visceral (kalazar).

The visceral leshimaniasis in Sudan is an endemic disease with sporadic out breaks and is considered an important socio-economic

problem. Clinically the cutaneous form is ulcerating granuloma of the skin. The visceral form is characterized by chronicity regular fever, enlargement of the spleen and often the liver and the presence of parasites in other organs. Donkeys are exposed to infection and play significant role in the epidemiology of the disease (Sharief, 1995).

2.6.3 Sarcoid

Sarcoid is the most common skin tumor. Equine sarcoids are locally invasive fibroblastic skin tumors that do not metastase . Substantial evidence supports a viral cause and possibly spread by flying insects. The condition is not highly contagious although the incidence of the disease is probably higher when donkeys are kept together. The lesions vary in appearance from apparently well defined nodules to rapidly growing ulcerated plaques with varying shapes (Reid *et al*., 1994). In Sudan the disease is mentioned as skin wart in the statistical Bulletin for animal Resources(2000).

2.6.4 Lameness

It is a common problem in donkeys. Most lameness are due to foot infection strained tendon sometimes occur in donkeys and injuries to joints can result in bruising and swelling.

The knees and fetlocks are being frequently affected which result in laminess. In Sudan it is frequently encountered in veterinary clinical (Hamid *et al*., 1998; Aradiab and Abbas, 1985).

2.6.5 Helminths infection in donkeys:

Equides are host to more than 75 species of helminths belonging to 28 genera of nematodes, 5 species of trematodes, and 4 species of cestodes (Krecek *et al*., 1987).

Heavy internal parasites burden can adversely affect the health of donkeys particularly when it is called upon to work and is often the case when under stress (Krecek et al 1987) .

Nematodes are the most important group of internal parasites. Most of nematodes have similar life cycle. Female of nematodes live in the donkeys intestine. The nematodes lay eggs which pass out in the manure and when the environment gets warm and moist the eggs hatch.

Some of the more common nematodes are ascarids, strongyles such as the lung worm and oxyuris. Ascarids are primarily a problem in young donkeys, a donkey usually develops resistance to ascarids at two months of age then they carry small number of it but in the foal ascarids may cause weight loss, develop rough hair coat, cough and produce

nasal discharge. Sometimes when dewormed the dead worms may pack into a large ball and obstruct the foals intestine. They cause colic and even death, so that a foal must be dewormed at 6 months of age to avoid worm accumulation in the intestine .

2.6.5.1 Strongylosis infection (Red worm infection)

Strongylosis is a common disease of horses. It occurs throughout the world and causes deaths when control measures are neglected.

Strongylus vulgaris is the most important parasite that causes vermenous arteritis and colic. *Strongylus edentates* and *S. equinus* also occur with lesser consequences (Blood *et al.*. 1983), these parasites migrate in the circulation and vital organs and cause severe damage that is fatal in some instances (Pandey and Eysker, 1989).

In Sudan the diseases are frequently reported in the Statistical Bulletin for Animal Resources (2000)and Hamid *et al*. (1998).

2.6.5.2 Lungworms

Donkeys are the natural hosts for lung worms and as such do not show obvious signs of disease when infected. The incidence of donkey infestation in Sudan is well known. (Seri.et al 2004) . Mules are reported to be relatively unaffected by lungworm infestation, similar to donkeys.

Horses however, may be severely affected exhibiting coughing and wheezing. Lungworms should be suspected if horses are pastured with donkeys or mules. Definitive diagnosis is made by demonstrating the presence of Dictyocaulus arnefeldi larvae in fresh faeces (Mathews and Taylor, 2005).

2.7 Skin disorders of donkeys

The donkey has for centuries been regarded as a robust and willing servant of man and most veterinarians accept that as a species it suffers rather fewer skin diseases than most other domestic animals. However this may be more in perception than in actuality because the donkeys tend to show few signs at cutaneous discomfort. The skin of the donkeys is well adapted to rigors of direct sunlight and extremes of heat (Kottenbelt, 2005)

The major syndroms of donkeys dermatology that might be encountered in practice include:

1. Pruritis , Nodular skin disease , Alopecia ,Moist/exudative dermatosis and Dry dermatosis (Flaking and scaling)(Knottenbelt ,2005)

2.8 Some studies on donkey parasites in the world

Ayele et al(2003) studied prevalence of gastrointestinal parasites of donkeys in Dugda Bora Distreict, Ethiopia. Their results showed 100% *Strongyle,* 50% *Parascaris equorum*, 4.5% *Anaplocephala sp*, 6% *Gastrodiscus agytiacus*, 3% *Oxuris equi a*nd 1.5% *Fasciola spp*. Eighty one point seven percent (81.7%) of donkey samples were severely infected, 8.3% were heavily infected, 23.8% and 6.2% were modratly and mildy infected respectly. The highest faecal egg count was in the long rainy season and the lowest worm egg count in the long dry season.

Yoseph and Smith (2005) studied theseasonal variation in parasites burden and body condition of working donkeys in West Shewa and East Shewa Region of Ethiopia. Their survey was conducted in donkeys arriving to market in 3 localities. Their result was: that the level of the helminthes infection closely followed the rainfall pattern begin lowest at 956 eggs per gram of faeces at the end of long dry season and the highest in the middle of the long wet season .Seasonal variation in body condition score reflect the temporal change in the worm burden and the availability of forage .It was suggested that body condition score of 3 or less could be used as simple means of identifying Donkeys that require

the Rapentic treatment with anthelmintic. It proposed that economically viable strategic control of helminths in the donkey could be achieved by administrating single annual dose of ivermectin as an anthelmintic at the start of the long rainy season. This may allow donkeys to recover their body condition when available forage is most abundant and nutritious. It is al so recommended that protein – rich feed supplementation be provided during the latter part of the dry season .This may help maintain body condition when forage is scarce and the heminths challenge is at its lowest.

Sonja *et al*. (2000) studied prevalence and biodiversity of helminth parasites in donkeys from South Africa. Samples were collected from the stomach, small intestine, cecum, ventral colon, dorsal colon, descending colon and cranial mesenteric artery.Their result revaeled the present of 15 genera and 29 species of helminths of nematodes from families, *Habronematidae, Onchocercidae, Oxyuridae* and *Strongylidae* and one (1) species of trematode from the *Paramphisto-matidae* in addition to two species of *Oestrid* fly larvae in the *Gastrophiliridae*. The most abundant group was the *cyathostomes 'small stronlyes"* and of the *cyathostomum Montgomery, cylic ocyclus spp* and

cylicostephanus minutes were the most numerous. The most prevalent *Cyathostomes* were *C. montgomeryi* and *cylicocyclus sip., Strongilus vulgaris* was the most abundant and prevalent as large *strongyle spp.* The occurrence of small *Strongyle spp.* and their prevalence in this study were compared with three other studies of the donkeys in Africa *(Pandy et al. 1989 ,1992)*

Wells *et al.* (1998) studied the helminths level of working donkeys kept under different management systems in the Moretele, South Africa. Faecal samples were collected from 93 donkeys in the study once a month for 14 months and were analyzed for nematode and trematode eggs and cultured to produce the third stage larvae for the identification of the nematode spp. Final comparison between management system subgroups, as well as between areas, age group and sexes were made. Four management systems were identified , the first system consisted of donkeys which were kept in small yard at all times. They were fed hay but no supplementary food , the second system consisted of donkeys which were allowed to roam freely around the village most of the time and rounded up and held in an enclosure when needed for work. the third system was identical to the second system except that the donkeys were

given supplementary food during winter . The fourth system was only found in the one area where each owner collects them into an enclosure prior to working. *Helminths* species composition and faecal egg count numbers differed between these four systems. The main difference noted was that donkeys from management systems (1) showed significantly higher number of *strongyle e*ggs and percentage of the s*trongyle* larvae. Management system (2) had higher stronyloides mean egg count and prevalence than the other groups. *Parascaris equirum and Gastrodiscus eqyptiation* egg count differed between all four systems. their result showed differences in the number and species of helminths in donkeys kept under the four management systems, suggestions are made as to which management system would facilitate reduction of helminth parasites in the animals. Although supplementary feeding in Moretele though fairly rare, it would seem that donkeys which do have access to better food resources have lower egg counts than donkeys on limited grazing.

Mushi *et al*. (2003) investigated the seasonal fluctuation of parasitic infection in donkeys in Oodi Village Kgatlong District Botswana. During the period March to September 2000, the study was

conducted in Twelve adult donkeys were randomly selected from a farmer with a herd of 15 donkeys. Monthly visits were made to the farmer when the donkeys were examined for parasites. The only ecto-parasites recovered from the donkeys were instars of various tick species. The most prevalent tick was *Rhipicephalus evertsi evertsi* (98.4%), followed by *Amblyoma hebraeum* and *Hyalomma* spp.

The only haemoparasite seen on microscopy was *Babesia equi* at low parasitaemia in (26.8%) of the donkeys. However no clinical babesiosis was evident. The coprological examination showed the presence of strongyle eggs in modrate numbers, very low number of coccidian oocysts were also found in the faecal samples. High tick number and worm egg counts coincided with the warm, wet months in contrast to the low numbers recovered during the cold, dry months. An interview conducted by owners showed that supplementary feeding was therefore, recommended specially during the winter month when grazing is poor.

Mukble *et al*. (2000) studied prevalence of hydatidosis among donkeys in Jordan . One hundred and thirty donkeys, aged between 5 months and 14 years of age were necropsied between November 1997 –

May 1999, of these animals 16.9% had hydatid cyst in either their lungs and\or livers. No donkeys of 3 years of age or less were infected, whereas 33.3% aged 4 years or greater were infected. The Intensity of infection increased with age in a linear fashion, the prevalence also increased with age approaching prevalence of 1 in the oldest animals. This implied there was minimal regulation of the parasites population by intermediate host immunity, the number of cysts in the donkeys was increased at rate of 0.48 cysts per year from 0.054 infection. The frequency distribution was highly aggregated, consistent with negative binomial distribution indicating infection of donkeys was not random.

Pankey *et al*. (1992) reported on epidemological observation on stomach worms of donkeys in Morocco. Over two consecutive years, weekly examination for the presence on nematodes were conducted on 185 stomachs from donkeys originated mainly from the Rabat Casablanca and Setta regions of Morocco. All the animals except one, were infected by at least one of four helminth species, *Trichostrongylus axei* was found in 93.5%, *Habronema muscae* in 89.7%, *Aronema majur*

in 85.4% and *Draschia megastoma* in 1.1% of donkeys. Most animals were infected by two(23.8%,) or three(71.9%) species.

High burdens of *T. axei* were observed in the winter of both years and in the mid summer of the second year peak burdens of *habronema* were found at various times throughout both years; there were more adult *H. majus* than *H. muscae*. The periods of peak levels of infection by these parasites were related to environmental condition suitable for the development and survival of infective larvae of *T. axei* and for the build up of muscid fly vectors of *Habronema* and *Drachia spp*.

Beelitz *et al .(1996)* studied about endoparasites of donkeys and horses kept in communal housing in Upper Pavaria, their species spectrum and incidence. In this epidemiological study of endoparasites 37 donkeys and 23 horses were included, which were examined microscopically in regular intervals over 15 month period. The animal derived from ten farms in which regular prophylactic treatment with anthelmintics had been practiced at least for two years before this investigation. This study revealed the presence of at least twelve parasites species in donkeys *Emeria leukart, Gastrophilus intestinalis*, *Fasciola hepatica, Anoplocephala perfoliata, Strongyloides westeri, Parascaris*

equorum, Dictyocaulus arnfieldi, Trichostrongylus axei and small *strongyles* sp. At least four parasites species were established in horses *E. leuckarti, G. intestinalis, P. equorum* and small *strongyles sp*

Infection with *E. leuckarti, S. westeri* and *P. equorum* were encountered in foals only and with *D. arnfieldi* in 16.2% of the donkeys, incidence for small *strongylus* sp. were highest exhibiting neither age and farm dependent nor host species dependent difference and amounting to 91.9% in donkeys and 86.6% in horses considering all donkeys and horses, 35.1% and 43.5% respectively were infected at least with two and 29.7%, 4.3% respectively with three parasites species. The species composition of the endoparasites fauna was always dominated by small *strongyles*. This field of study indicates that the species composition of the endoparasites fauna in comparison to former investigations has not or only slightly changed in spite of widespread use of broad spectrum anthelminics for years, in addition it has to be considered that donkeys continue to act as reservoir of *D. arnifieldi* and as source of infection for horses.

Seri *et al*. (2004) reported on the prevelance of gastrointestinal nematodes of donkeys in Khartoum State, Sudan. They study 1200

donkey during a complete year (one hundred per month) were examined parasitically for presence of helminth parasitiology by assessment of faecal egg and culture identification of larvae.

Generally 70% of examined animals harboured a parasitical infection. Six nematodes genera were encountered in the donkey, among them *Dictycoulus arnfield* 70.5% reported for the first time. In Sudanese donkey*Stronglyes spp.* 35-8%, *Cyathosome spp* 38-6.7, *Parascaris equarum* 10.7% and *Trichstrongylus axie* 12% and *Strongyloides westeri* 34.1. the incidence of infection with one species 53.2 was found higher than that of mixed infection 46.7. According to severity of infection, 58.6% of the infected animal showed mild infection while 21.9 and 19.0% of them showed moderate and severe infection respectively.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Samples collection

3.1.1 Blood examination

From 100 randomly selected donkeys, peripheral blood smears were prepared, preparations of thin blood smears were done according to (McCosker, 1975), the edge of the donkeys ears were cleaned with methanol-absorbed piece of cotton wool, allowed to dry and then pricked with a sterile needle. The first drop of blood was collected on a clean grease-free microscopic slide and smeared very carefully, from each donkey. Two blood films were made. Each slide was marked on one edge of the blood film with a pencil. The slides were immediately stored in slide box to prevent contamination with dust.

A few milleliters of blood from the jugular vein of each donkey were also taken in EDTA- containing vaccutainer tube and placed in the ice box and brought as soon as possible to the laboratory for inoculation into mice.

Collected blood smears were fixed in absolute methyl alcohol for three to four (3-4) minutes then stained with Giemsa stain.

The Giemsa solution was diluted in buffered distilled water (BDW) at pH (7.2). Ten percent of Giemsa stain solution was freshly prepared for each group of samples i.e. 10 cc of the stock solution of Giemsa were added to 90 cc of the BDW and allowed to stay for at least half an hour before use.

Before dilution, the stock solution of Giemsa was filtered using Whatmann filter paper No. (1) to avoid stain particles depositing on the stained slide. The air dried fixed blood films were them immersed in the staining jar to stain for 45 minutes. They were then removed, washed carefully with distilled buffer water pH 7.2 and allowed to dry.

The smears were scanned for intraerythrocytic stage of *Babesia spp* or other parasites under Leitz (Dialux 20) research microscope using oil immersion objectives (10x100), each slide was scanned for about 20 minutes.

Blood collected in vaccutainers was injected into experimental animals (mice) in their peritonial cavity and every three days smears

were prepared from a cut of the tail of mice and fixed and stained and examined for blood parasites

3.2 Faecal samples

3.2.1 Collection of faeces

A total of 100 faecal samples were collected from individual donkeys; fresh samples were collected in plastic bags using hand gloves from the rectum of the donkeys; the plastic bags were filled to the top and tied well so as to exclude air and hence diminish the rate of development and hatching of eggs; then the bags were labeled and taken to the laboratory as soon as possible for examination and cultivation for parasite larvae.

3.2.2 The Floatation method

One hundred faecal samples were examined using sodium chloride as floatation solution, 2 or 3 gram of faeces were ground in a mortar and pestle and placed in a tube. Saturated Sodium chloride was added to the tube which was filled to the top with the same faecal mixture till it made a convex meniscus.

A microscopal slide was inverted over the top of the tube, after 15 minutes the slides was quickly removed and covered with cover slip and examined under a microscope for parasite eggs or larvae.

3.2.3 Culture method

3.2.3.1 Faecal culture for third stage infective larvae

Faecal samples from individual donkeys were cultured, the third stage infective larvae were obtained by using a similar method to what has been described by (Robert, 1950).

Firstly 20 gram of faeces were ground using pestle and morter and wrapped in piece of guaze and then suspended in a closed marmalade jar containing a small amount of water to provide the media with moisture which is important for hatching of eggs to release larvae.

3.2.3.2 Recovery of larvae

Living L3 larvae were harvested 7-10 days after culturing using Baerman technique (Dunn, 1978). The Baerman apparatus is composed of funnel with a rubber tube attached at its bottom end and closed with a clamp or spring clip. The funnel is fixed to a stand. The cultured faeces were wrapped in a double layered gauze and placed in the funnel. Water was added until it covered the faeces in the gauze, the apparatus was left

for 2-3 hours most of the larvae would sink to the bottom of the funnel, specially when the apparatus was allowed to stand overnight, as described in many author (Anon, 1977; Dunn 1978; Anon, 1986).

This would result in the death of most larvae under warm conditions. Such dead larvae assume a curved posture which makes differentiation by micrometry extremely difficult. The first ml of water at the bottom of the funnel was drawn off in a test tube and kept in a refrigerator (4c) to slow down the movement of the larvae. One drop of the culture fluid was placed on a slide after adding formaldehyde (formalin) to kill the larvae, a cover slip was placed over the slide. It was examined microscopically and from each sample almost all larvae were identified and measured. The species identity of the L3 larvae was established using the following criteria: presence of the sheath, body length, length of the oesophagus, shape and anterior structure of the head, number and shape of the intestinal cells, relative size and shape of the sheath tail and the shape of the larval tail (Dunn, 1978; Anon, 1977, 1986) larval dimensions were measured using a graticule in the eyepiece micrometer fitted to the standard microscope.

3.2.3.2 Egg count method

A total of 100 faecal samples was examined using Mc master egg counting technique to demonstrate the presence and number of egg per gram of faeces. This quantitative method was described by (Soulsby, 1986).

Two grams of donkey faeces were dissolved in 10 ml of tap water and the mixture was ground using pestle and morter, then 50ml of saturated sodium chloride was added, the mixture was then strained through fine sieve and the residue was pressed out in a plastic cap. A sample was stirred well in order to obtain complete homogenous distribution of the eggs in the mixture, the solution was then drawn by a Pasteur pipette and was run in the two counting chambers of the Mc master slide. The slide was left for a few minutes to allow eggs to float up and the sample was examined under the low power of the microscope, the eggs in the two chambers were counted and the number of eggs per gram of faeces was obtained according to the following formula:

$$\text{The total helminth egg count} = \frac{\text{Total of the egg in two chamber} \times 100}{2}$$

3.2.3.3 Ticks on the donkeys:

Search for ticks in the donkeys revealed the absence of the ticks.

The prevalence of infection was not significantly differed between the three localities .

CHAPTER FOUR

RESULTS

4.1 Faecal examination

Examination of 100 faecal samples from donkeys originating from Khartoum, Omdurman and Khartoum North City. Revealed presence of numerous parasite eggs.

4.2 Faecal culture

Faecal culture of samples from donkeys from three localities revealed presence of third larval stages (L3) of nematodes.

Table (2): No. of infected and non-infected animals in Khartoum State

Local	No. of animals	Infected	Non infected	% infected
Omdurman	35	23	12	65.5
Khartoum North City	35	22	13	60.8
Khartoum	30	17	13	56.6
Total	100	62	38	100

Table (3): Egg count in Khartoum State

No.	Khartoum (egg/gram)	Omdurman (egg/gram)	Khartoum north city (egg/gram)
1	150	250	50
2	200	200	350
3	150	150	500
4	350	100	450
5	200	300	50
6	150	250	150
7	150	200	550
8	50	100	300
9	50	100	250
10	150	100	50

No blood parasites had been detected upon blood examination , like wise ,the examination of the study animals showed the absence of adult or developmental stages of ticks .

The identified Nematode eggs were depicted in fig (1) and (2). Fig (3)through fig (6) illustrated the third stage larvae of *Strongyles vulgaris ,Strongyles edentatus ,Strongyles equinus and Trichonema spp* respectively

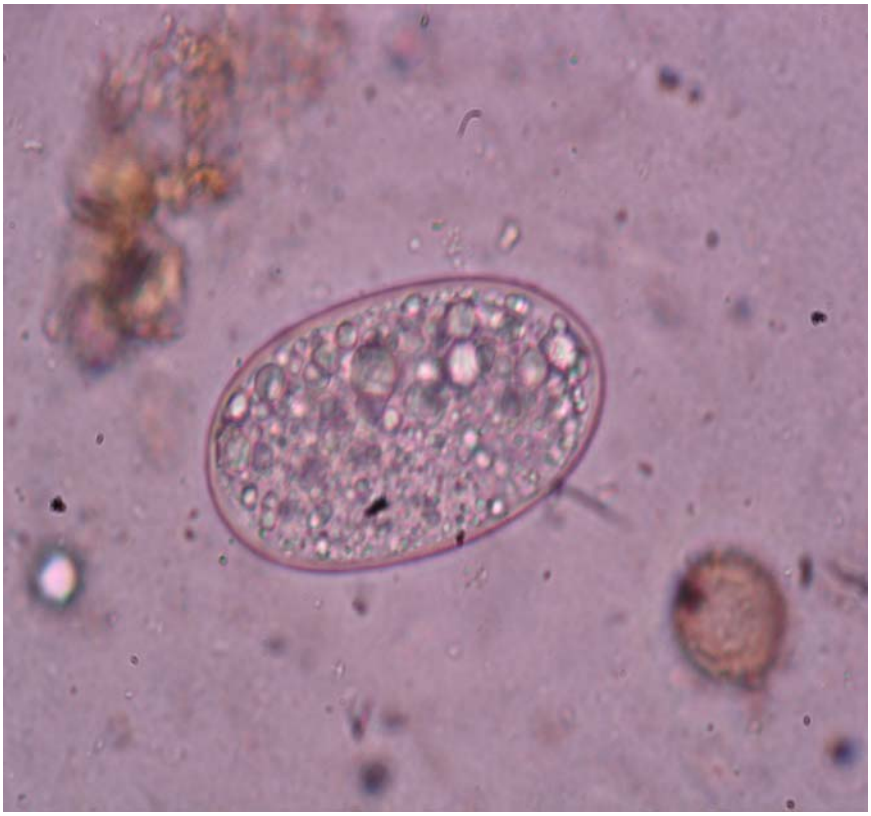

Fig. (1): Egg of a Nematode worm, undifferentiated

Fig. (2): Egg of a Nematode worm (undifferentiated), note the encapsulation

Fig. (3): *Strongylus vulgaris* larva after release from the egg

Intestinal cells are 28-32 rectangular cells

Fig. (4): *Strongylus edentatus* larva
had 20 intestinal cells

Figure (5): *Strongylus equinus* larva
had 16 intestinal cells

Fig. (6): *Cyathostoma* (*Trichonema*) spp.

had 8 triangular intestinal cells

CHAPTER FIVE

DISCUSSION AND CONCLUSION

Donkeys especially if they look apparently health are generally reputed to suffer from very few diseases. This study was formulated with this idea which stipulates that if any diseases were found, they would, in any case, be few.

It was found convenient to subject working donkeys in Khartoum to examination of their blood and faecal material. Only working donkeys who are apparently healthy were selected with the assumption that they might be hiding some infection which are in apparent.

Of course there are donkeys which are roaming around neglected after long use for years and having lost their stamina and outlined their usefulness. They are usually found in the outskirts of towns, deprived of any care, ownerless and homeless. Such donkeys would have been most suitable as subjects for study. However their availability in statistical numbers was in doubt. It may take a considerable time to collect a satisfactory number to examine for the

diseases they suffer from. It would even be most difficult to ascertain how they contracted their diseases in the first place.

Therefore it was found more sound and logical to focus on the working donkeys only owing to their availability and ease of access to them.

Blood parasites such as babesia, trypanosomes and theileria would lower the stamina of the donkeys and limit their usefulness as draft and carrier animal. Their owners rush to veterinary clinics to get them treated to regain their health, as the owners depend on them for their daily livelihood. Therefore it was found that most of the donkeys examined were found in their best health.

Therefore, it is not surprising that most donkeys failed to show blood parasites. It was also observed that all the donkeys examined failed to carry ticks of any kind, reflecting the care they receive from their owners.

Round worms constituted most of the parasites found in the donkeys, strongyles were most prominent. An effort was made to further identify them through allowing the worm eggs to develop into larvae which carry the features of the parent worms was made. Only strongyles

and trichonema worms were found judging from the morphology of the larvae only. These criteria were based on the internal structure and extremities of the larvae. It would have been more accurate if these larvae were allowed to develop to the stage of (L3) larvae.

The most important finding is that all donkeys carried nematode eggs and larvae without showing any ill health. Their egg counts varied and were significantly high but they seem to tolerate these worms without any ill effect.

This confirms that donkeys are extremely useful beasts of burden which do work with high efficiency and without significant disease problems.

The results obtained indicated high rates of helminths among the tested animals in three localities; this suggests that parasite infections are in wide distribution in Khartoum state and most nematodes are *Strongylus vulgaris. S. equines, S. edentatus* and *Cythostom spp*.

In spite of the numerous reports and studies on parasites affecting livestock in Sudan no investigation was made to the donkeys parasites.

From the above results we recommended the following:

1. To pay more attention to donkey nematodes in this State.

2. To conduct similar surveys for gastrointestinal helminths in donkeys from other areas in the Sudan.
3. To conduct more studies on the species reported for the first time in this country such as *Strongylus vulgaris , Strongylus edentatus , Strongylus equinus and Trichonema spp* ,and continue research to detect more species that may be present such as donkey parasites are numerous and world-wide distribution.
4. To observe areas and conditions in which donkeys are kept and try to minimize the risks of infection.
5. To draw attention to the donkeys and try to minimized inapparent losses such as inability to work due to gastrointestinal helminths.
6. This study used blood smear examination as the only technique. Some infection with blood parasites were missed due to the limitation of the technique. If possibly more sensitive technique such as the micro-haematocrit would have shown some blood parasites such as tryponasomes.
7. It is recommended that a serological survey should have been conducted to clarify the situation of circulating antibodies to different blood parasites in donkeys.

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